Letters to the Editor . . .

SURFACE PHAGOCYTOSIS

A hitherto unsuspected mechanism of phagocytic activity of major clinical interest is described by Wood[3] and his associates of Washington University School of Medicine, St. Louis, Missouri.

Phagocytes were obtained from peritoneal exudates of rats injected 24 hours previously with an aleuronat broth mixture. Approximately 90 per cent of the cells thus obtained were polymorphonuclear leucocytes and 10 per cent macrophages. Suspensions of thrice-washed phagocytes in gelatin-Locke's solution were mixed with washed type I pneumococci. No phagocytosis took place in this mixture when incubated in rotating glass tubes, as hanging drop preparations, or when spread on the surfaces of glass, paraffin, albumin or cellophane. This is in line with conventional theory which assumes that phagocytosis of encapsulated pneumococci is impossible except in the presence of type-specific opsonins.[2]

In contrast with these negative findings, highly active phagocytosis took place when the same mixture was brought into contact with filter paper, lens paper, cloth or fiberglas. From these observations it was concluded that both polymorphonuclear leucocytes and macrophages, when given access to a suitable surface, will phagocytize virulent pneumococci without the aid of an intermediary antibody or any other tissue factor. The St. Louis investigations suggest the use of the term "surface phagocytosis" for this new phenomenon, but without suggesting a definite theory as to the physical, chemical or biological factors involved.

In order to test possible clinical applications of this new phenomenon, small pieces of selected tissues taken from freshly killed rats were placed in the bottom of Petri dishes lined with moistened filter paper. A small drop of leucocyte-pneumococcus mixture was spread over each tissue surface. Each Petri dish was then sealed with Scotch tape, and incubated for one hour. Impression smears from the tissue surfaces demonstrated highly active phagocytosis on each tissue surface. Among the tissues tested were bronchial and tracheal epithelium, esophageal epithelium, the intima of both aorta and vena cava, pleura, pericardium, endocardium, peritoneum, mesentery, retina, muscle, and clotted plasma. When the same tissues had been previously boiled for 30 minutes the same phagocytic action took place.

The phagocyte-pneumococcus mixtures were then injected intrabronchially into (a) the lungs of normal rats, (b) the lungs removed from rats and perfused with gelatin-Locke's solution, and (c) rat lungs fixed for 24 hours in 10 per cent formalin and then washed for several days to remove the fixative. Each experiment was carried out at body temperatures. Sections cut from each type of preparation showed that both polymorphonuclear leucocytes and macrophages phagocytized pneumococcus in the alveoli within less than an hour. In the formalin-fixed lungs there was no possible source of opsonin.

Examination of the formalin-fixed lungs revealed that pneumococci were engulfed by the phagocytic cells in the larger bronchi as well as by those in the alveoli. Phagocytosis failed to occur, however, when the same leucocyte-pneumococcus mixtures were tested in capillary glass tubes of the same diameter as the bronchi. This suggests that the crucial factor in the phagocytic process is related in some way to the character of the bronchial surface, justifying the suggested terminology "surface phagocytosis."

Wood finds clinical confirmation of his findings in the abundant phagocytosis that takes place in the early stages of lobar pneumonia, before the appearance of specific antibodies.[1]

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ISOLATION OF Rh-HAPTEN

In 1944 it was shown by Belkin and Wiener[1] of the Jewish Hospital, Brooklyn, N. Y., that Rh-antigen is present quantitatively in the stromata of Rh-positive human red blood cells. They found that such stromata could be injected intramuscularly into human volunteers, without demonstrable toxic reactions. From this they suggested the clinical use of such stromata in attempted desensitization of Rh-negative mothers while carrying a Rh-positive fetus. The possibility of a successful application of this technique is increased by the isolation and purification of the Rh-antigen and hapten from such stromata, currently reported by Calvin[2] and his associates of the Department of Chemistry, University of California and the Department of Medicine, Stanford University.

Rh-positive erythrocytes centrifuged from citrated human blood were laked at 5°C by the addition of eight to ten volumes of distilled water. The stroma thus obtained was repeatedly washed by super-centrifugation and dried by lyophylization. Approximate yield 1-2 g. per 250 cc. r.b.c. The dry product thus obtained dissolved at pH 7-8 yielded a solution which completely inhibited Rh-agglutination by anti-Rh serum when tested in relatively high dilutions. Control tests with Rh-negative stromata gave negative results.

By a modification of the Jorpes[3] technique, this Rh-positive stroma was separated into two fractions. About 60 per cent of the stroma complex was precipitated by ultracentrifugation at pH 5.5 to 6.0. This "stromatin" fraction contained no demonstrable Rh-antigen. By changing the pH titer to 7-8 a second ultracentrifugable fraction was obtained. For this the California investigators propose the term "elinin." Elinin is a lipoprotein, and contains the Rh-antigen quantitatively.

Adopting a modification of the Landsteiner[4] technique a lipoidal fraction was extracted from elinin by the use of 3:1 alcohol-ether. This lipoid constitutes from 40 to 50 per cent of the elinin complex, and apparently contains the Rh-hapten quantitatively. If this lipoid has immunological properties similar to those of other known haptens, one would expect it to serve as an effective prophylactic and desensitizing agent in the prevention or treatment of erythroblastosis fetalis[5] and related toxemias. Clinical trials, however, have not yet been reported.

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